

SUPPLEMENTAL AMENDMENT UNDER 37 C.F.R. § 1.116 Attorney Docket No.: Q88078
Application No.: 10/536,768

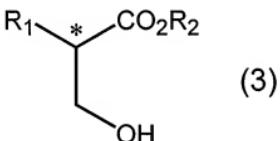
AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

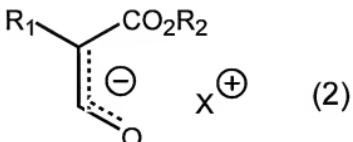
LISTING OF CLAIMS:

1.-6. (canceled).

7. (currently amended): A process for producing an optically active 3-hydroxypropionic ester derivative represented by the formula (3):



where R₁ represents an alkyl group having 2 to 10 carbon atoms, an optionally substituted aralkyl group having 5 to 15 carbon atoms, or an optionally substituted aryl group having 5 to 15 carbon atoms; R₂ represents an alkyl group having 1 to 10 carbon atoms, or an optionally substituted aralkyl group having 5 to 15 carbon atoms; and * represents an asymmetric carbon atom, characterized by subjecting a 2-formylacetic ester derivative represented by the formula (2):



where R_1 and R_2 are the same as described above; and X represents H, Li, Na or K, to the action of an enzymatic source capable of stereoselectively reducing the formyl group thereof,

wherein the R configuration of the derivative represented by the formula (3) is produced by using an enzymatic source which is derived from a microorganism of the genus of *Brettanomyces*, *Debaryomyces*, *Galactomyces*, *Ogataea*, *Pichia*, *Saccharomyopsis*, *Sporidiobolus*, *Sporobolomyces*, *Sterigmatomyces*, *Torulaspora*, *Trichosporon*, *Yamadazyma*, *Achromobacter*, *Cellulomonas*, *Devosia*, *Hafnia*, *Jensenia*, *Klebsiella*, *Micrococcus*, *Proteus*, or *Serratia*, and capable of R-selectively reducing the formyl group of the derivative represented by the formula (2); or

the S configuration of the derivative represented by the formula (3) is produced by using an enzymatic source which is derived from a microorganism of the genus of *Cystofilobasidium*, *Pichia*, *Torulaspora*, *Williopsis*, *Yarrowia*, *Devosia*, *Microbacterium*, or *Micrococcus* and capable of S-selectively reducing the formyl group of the derivative represented by the formula (2), and

recovering the optically active 3-hydroxypropionic ester derivative represented by the formula (3) after the stereoselective reduction.

8. (original): The process according to claim 7 wherein in the formulas (2) and (3), R_1 is an alkyl group having 2 to 10 carbon atoms or an optionally substituted aralkyl group having 5 to 15 carbon atoms.

9. (previously presented): The process according to claim 7 or 8 wherein the R configuration of the derivative represented by the formula (3) is produced by using, as the R-selective enzymatic source, an enzymatic source derived from a microorganism selected from the group consisting of *Brettanomyces anomalus*, *Debaryomyces nepalensis*, *Debaryomyces robertsiae*, *Galactomyces reessii*, *Ogataea minuta* var. *minuta*, *Pichia canadensis*, *Pichia silvicola*, *Pichia xylosa*, *Saccharomyopsis selenospora*, *Sporidiobolus johnsonii*, *Sporidiobolus salmonicolor*, *Sporobolomyces salmonicolor*, *Sterigmatomyces halophilus*, *Torulaspora delbrueckii*, *Trichosporon asteroides*, *Yamadazyma stipitis*, *Achromobacter xylosoxidans* subsp. *denitrificans*, *Cellulomonas fimi*, *Cellulomonas* sp., *Cellulomonas uda*, *Devosia riboflavina*, *Hafnia alvei*, *Jensenia canicuria*, *Klebsiella planticola*, *Micrococcus luteus*, *Proteus inconstans*, and *Serratia marcescens*

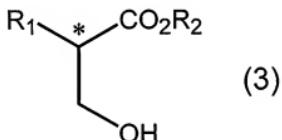
10. (previously presented): The process according to claim 7 or 8, wherein the R-selective enzymatic source is a cultured product of *Escherichia coli* HB101 (pNTDRG1)(FERM BP-08458), or *Escherichia coli* HB101 (pTSBG1)(FERM BP-7119); or a processed product thereof.

11. (currently amended): The process according to claim 7 or 8 wherein the S configuration of the derivative represented by the formula (3) is produced by using, as the S-selective enzymatic source, an enzymatic source derived from a microorganism selected from the group consisting of *Cystofilobasidium bisporidii*, *Pichia bispora*, *Torulaspora globosa*, *Williopsis saturnus* var. *mrakii*, *Williopsis saturnus* var. *saturnus*, *Yarrowia lipolytica*, *Devosia riboflavina*, *Microbacterium esteraromaticum*, and *Micrococcus luteus*.

12. (previously presented): The process according to claim 7 or 8, wherein the S-selective enzymatic source is a cultured product of *Escherichia coli* HB101 (pNTDRG1)(FERM BP-08458), or *Escherichia coli* HB101 (pTSBG1)(FERM BP-7119); or a processed product thereof.

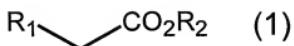
13.-16. (Canceled).

17. (currently amended): A process for producing an optically active 3-hydroxypropionic ester derivative represented by the formula (3):



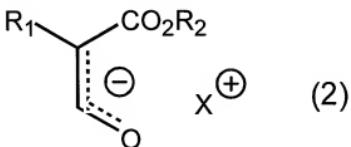
where R_1 represents an alkyl group having 2 to 10 carbon atoms, an optionally substituted aralkyl group having 5 to 15 carbon atoms, or an optionally substituted aryl group having 5 to 15 carbon atoms; and R_2 represents an alkyl group having 1 to 10 carbon atoms, or an optionally substituted aralkyl group having 5 to 15 carbon atoms[[;]]; and * represents an asymmetric carbon atom, characterized by comprising the steps of:

reacting an acetic ester derivative represented by the formula (1):



where R_1 and R_2 are the same as described above

with a base and a formic ester, thereby converting the acetic ester derivative into a 2-formylacetic ester derivative represented by the formula (2):



where R₁ and R₂ are the same as described above; and X represents H, Li, Na or K;

removing impurities from the reaction mixture into an organic layer formed by addition of an organic solvent and water thereto, while transferring/dissolving the derivative represented by the formula (2) into a resulting aqueous layer; and

stereoselectively reducing the derivative represented by the formula (2) by use of an enzymatic source capable of stereoselectively reducing the formyl group of the derivative represented by the formula (2), thereby obtaining the optically active 3-hydroxypropionic ester derivative represented by the formula (3);

wherein the enzymatic source is derived from a microorganism belonging to the genus *Brettanomyces*, *Debaryomyces*, *Galactomyces*, *Ogataea*, *Pichia*, *Saccharomyces*, *Sporidiobolus*, *Sporobolomyces*, *Sterigmatomyces*, *Torulaspora*, *Trichosporon*, *Yamadazyma*, *Achromobacter*, *Cellulomonas*, *Devsia*, *Hafnia*, *Jensenia*, *Klebsiella*, *Proteus*, *Serratia*, *Cystofilobasidium*, *Williopsis*, *Yarrowia*, *Microbacterium*, or *Micrococcus*, and recovering the optically active 3-hydroxypropionic ester derivative represented by the formula (3) after the stereoselective reduction.

18. (original): The process according to claim 17 wherein, in the formulas (1), (2), and (3), R₁ is an alkyl group having 2 to 10 carbon atoms or an optionally substituted aralkyl group having 5 to 15 carbon atoms.

19. (original): The process according to claim 17 or 18 wherein, in the formulas (1), (2) and (3), R₂ is an alkyl group having 1 to 10 carbon atoms.

20. (canceled).

21. (currently amended): The process according to claim 1720 wherein the R configuration of the derivative represented by the formula (3) is produced by using an enzymatic source which is derived from a microorganism belonging to the genus of *Brettanomyces*, *Debaryomyces*, *Galactomyces*, *Ogataea*, *Pichia*, *Saccharomyopsis*, *Sporidiobolus*, *Sporobolomyces*, *Sterigmatomyces*, *Torulaspora*, *Trichosporon*, *Yamadazyma*, *Achromobacter*, *Cellulomonas*, *Devsia*, *Hafnia*, *Jensenia*, *Klebsiella*, *Micrococcus*, *Proteus*, or *Serratia* and capable of R-selectively reducing the formyl group of the derivative represented by the formula (2).

22. (previously presented): The process according to claim 21 wherein the enzymatic source capable of R-selective reduction is derived from a microorganism selected from the group consisting of *Brettanomyces anomalus*, *Debaryomyces nepalensis*, *Debaryomyces robertsiae*, *Galactomyces reessii*, *Ogataea minuta* var. *minuta*, *Pichia canadensis*, *Pichia silvicola*, *Pichia xylosa*, *Saccharomyopsis selenospora*, *Sporidiobolus johnsonii*, *Sporidiobolus salmonicolor*,

Sporobolomyces salmonicolor, *Sterigmatomyces halophilus*, *Torulaspora delbrueckii*,
Trichosporon asteroides, *Yamadazyma stipitis*, *Achromobacter xylosoxidans* subsp. *denitrificans*,
Cellulomonas fimi, *Cellulomonas* sp., *Cellulomonas uda*, *Deeosia riboflavina*, *Hafnia alvei*,
Jensenia canicururia, *Klebsiella planticola*, *Micrococcus luteus*, *Proteus inconstans*, and *Serratia*
marcescens.

23. (previously presented): The process according to claim 22 wherein the enzymatic source capable of R-selective reduction is a culture product of *Escherichia coli* HB101 (pNTDRG1)(FERM BP-08458), or *Escherichia coli* HB101 (pTSBG1)(FERM BP-7119); or a processed product thereof.

24. (currently amended): The process according to claim 1720 wherein the S configuration of the derivative represented by the formula (3) is produced by using an enzymatic source which is derived from a microorganism belonging to the genus of *Cystofilobasidium*, *Pichia*, *Torulaspora*, *Williopsis*, *Yarrowia*–*Deeosia*, *Microbacterium*, or *Micrococcus* and capable of S-selectively reducing the formyl group of the derivative represented by the formula (2).

25. (currently amended): The process according to claim 24 wherein the enzymatic source capable of S-selectively reducing the formyl group of the derivative represented by the formula (2) is derived from a microorganism selected from the group consisting of *Cystofilobasidium bisporidii*, *Pichia bispora*, *Torulaspora globosa*, *Williopsis saturnus* var.

mrakii, *Williopsis saturnus* var. *saturnus*, *Yarrowia lipolytica*; *Devsia riboflavina*,
Microbacterium esteraromaticum and *Micrococcus luteus*.

26. (previously presented): The process according to claim 24 or 25 wherein the enzymatic source capable of S-selectively reducing the formyl group of the derivative represented by the formula (2) is a cultured product of *Escherichia coli* HB101 (pNTDRG1)(FERM BP-08458), or *Escherichia coli* HB101 (pTSBG1)(FERM BP-7119); or a processed product thereof.

27. to 31. (canceled).